

1 ONLINE SUPPLEMENTARY MATERIAL

2 Of the 15 single nucleotide disagreements between our *de novo* NR1 sequence
3 and the GenBank R100 sequence (Table 4), nine arise from an eleventh QQP repeat near
4 the carboxy-terminus of the transfer protein TraD in the *de novo* NR1 sequence. TraD
5 proteins, which function as inner membrane ATPases, of other *E. coli* and *Salmonella*
6 plasmids (e.g., NC_005327, NC_006671, NC_006855) have a variable number of QQP
7 repeats, suggesting that this is a real polymorphism.

8 Another disagreement is a single base change within codon 301 of the mating pair
9 stabilization protein TraN. The *de novo* NR1 encodes glutamate (GAA), but the GenBank
10 R100 encodes a stop codon (TAA). The *traN* gene of R100-1, a derepressed variant of
11 R100 with a mutation in *finO*, has also been sequenced and has a glutamate at the
12 corresponding position (5), supporting the accuracy of our NR1 sequence at this position.
13 NR1 is conjugative (unpublished observations), which further supports the functionality
14 of TraN.

15 The remaining five disagreements are within a ~1400 bp region of *IS10*-R of
16 *Tn10*, which contains the functional transposase of this composite transposon (4), and the
17 3' flanking sequence. Three are synonymous third base substitutions in the *IS10*-R
18 transposase, two of which (NR1 ATCile:R100 ATTile; NR1 CGGarg:R100 CGAarg) are
19 identical to the corresponding positions in *IS10*-L as previously reported (4). The third
20 (NR1 GCAala:R100 GCCala) is identical to the corresponding position in the previously
21 reported *IS10*-R (4), whereas the GenBank R100 sequence agrees with *IS10*-L (4). A
22 fourth disagreement in the ~1400 bp region is within *yefA*, a hypothetical protein near
23 *IS10*-R but outside *Tn10*. This conservative substitution (NR1 TTCphe:R100 TACTyr)

24 was previously observed in *yefA* of plasmids F and R6-5 (3). The last disagreement in the
25 ~1400 bp region is in the plasmid SOS inhibition protein PsiB, which is proposed to
26 prevent induction of the bacterial SOS response during conjugative transfer in order to
27 protect single-stranded plasmid DNA (1). The NR1 sequence has a single nucleotide
28 deletion in codon 127 of PsiB, resulting in a frameshift and premature termination. This
29 *psiB* polymorphism is in the C-terminal region, which when deleted in other examples of
30 this gene does not affect the protein's activity (2).

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31 **REFERENCES:**

- 32
- 33 1. **Bagdasarian, M., A. Bailone, J. F. Angulo, P. Scholz, M. Bagdasarian, and R.**
- 34 **Devoret.** 1992. PsiB, an anti-SOS protein, is transiently expressed by the F sex
- 35 factor during its transmission to an *Escherichia coli* K-12 recipient. Mol.
- 36 Microbiol. **6**:885-893.
- 37 2. **Bailone, A., A. Backman, S. Sommer, J. Celerier, M. M. Bagdasarian, M.**
- 38 **Bagdasarian, and R. Devoret.** 1988. PsiB polypeptide prevents activation of
- 39 RecA protein in *Escherichia coli*. Mol. Gen. Genet. **214**:389-395.
- 40 3. **Dutreix, M., A. Backman, J. Celerier, M. M. Bagdasarian, S. Sommer, A.**
- 41 **Bailone, R. Devoret, and M. Bagdasarian.** 1988. Identification of *psiB* genes of
- 42 plasmids F and R6-5. Molecular basis for *psiB* enhanced expression in plasmid
- 43 R6-5. Nucleic Acids Res. **16**:10669-10679.
- 44 4. **Halling, S. M., R. W. Simons, J. C. Way, R. B. Walsh, and N. Kleckner.** 1982.
- 45 DNA sequence organization of IS10-right of Tn10 and comparison with IS10-left.
- 46 Proc. Natl. Acad. Sci. USA **79**:2608-2612.
- 47 5. **Klimke, W. A., and L. S. Frost.** 1998. Genetic analysis of the role of the transfer
- 48 gene, *traN*, of the F and R100-1 plasmids in mating pair stabilization during
- 49 conjugation. J. Bacteriol. **180**:4036-4043.

52 Table 4. Disagreements between *de novo* NR1 sequence and R100 reference sequence
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Number of disagreements	Location	Type
9	TraD	additional QQP repeat in NR1 sequence
1	TraN	NR1 GAA glu: R100 TAA stop
3	IS10-R transposase	synonymous 3 rd base substitutions
1	yefA, hypothetical protein	conservative amino acid substitution
1	PsiB	1 nt deletion resulting in frameshift and premature termination

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